Appendix B

The Influence of Formulation Composition on the Stability of Freeze-Dried Equine Synovial Fluid

Summary

A study was carried out to compare the solid-state stability profile of five different formulations of equine synovial fluid at -20° C, and -80° C, and 5° C over a period of eight weeks. A mucin clot test was used as a measure of product quality. Although the variability of the mucin clot test was a major limitation of this study, formulations containing 10% Mannitol and 10% Maltodextrin provided the best results as measured by clot quality over the course of the study. Polysorbate 80 and human serum albumin seems to offer no advantage. A formulation containing Mannitol and Glycine was nearly equivalent to the formulations containing Maltodextrin. The stability data did not support a clear conclusion as to whether there is a significant loss of activity during storage at either 5° C or -20° C. Surprisingly, there appears to be a loss of clot quality with time during storage at -80° C. There is significant loss of activity during storage of the reconstituted product at room temperature.

Objective

The purpose of this experiment was to compare the solid-state stability of five different formulations of equine synovial fluid over an eight-week interval at temperatures of -80° C, -20° C, and 5° C in order to identify the most promising formulation for further study.

Experimental

Materials

Equine synovial fluid (Lot 121399) was used as received from Dr. Paul Christofferson of Equine Bio-Tech Inc. Maltodextrin (M500) was from Grain Processing Corporation, Muscatine, IA. Mannitol and Glycine were analytical grade material from Mallinckrodt (Paris, KY). Polyexyethylene sorbitan monooleate (Tween 80) was from Sigma Scientific (St. Louis, MO). Human serum albumin was a gift from Bayer Corporation (Berkeley, CA).

Methods

Preparation of Formulations

The stabilizer systems used in this study were as follows:

Formulation # 1:

10% Mannitol
10% Maltodextrin